

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings of claims in the application:

**Listing of Claims:**

1-9. (Canceled)

10. (Currently Amended) An antibody, comprising:

~~at least one point~~ points of conjugation for a cytotoxic or cytostatic agent, wherein  
[[the ]] at least one point of conjugation for the cytotoxic or cytostatic agent on the antibody can  
be readily assigned, and wherein less than all possible points[[ are]] of conjugation are available  
for conjugation to the cytotoxic or cytostatic agent.

11. (Currently Amended) The antibody of claim 10, wherein the points at  
least one readily-assignable point of conjugation ~~are~~ is an interchain thiols thiol, and the antibody  
comprises at least one interchain disulfide bond.

12. (Canceled)

13. (Currently Amended) The antibody of claim[[ 10]] 11, wherein the  
antibody has four points of conjugation, and wherein the point of conjugation is antibody has a  
configuration of an antibody species selected from at least one of the group consisting of species  
4A, 4B, 4C, 4D, 4E and through 4F of Figure 1.

14-33. (Canceled)

34. (Currently Amended) A method of reducing and conjugating a drug to  
an antibody resulting in selectivity in the placement of the drug, comprising:

fully reducing the antibody with a reducing agent;

treating the fully reduced antibody with limiting amounts of a reoxidizing agent to  
reform at least one interchain disulfide bond of the antibody, such that at least two interchain  
thiols remain; and

conjugating the drug to ~~the one~~ interchain thiols thiol.

35. **(Original)** The method of claim 34, wherein the reoxidizing agent is 5,5'-dithio-bis-2-nitrobenzoic acid, 4,4'-dithiodipyridine, 2,2'-dithiodipyridine, sodium tetrathionate or iodosobenzoic acid.

36. **(Currently Amended)** The method of claim ~~34~~ 35, wherein the drug is a cytotoxic or cytostatic agent or an immunosuppressive agent.

37. **(Currently Amended)** The method of claim ~~34~~ 36, wherein the cytotoxic or cytostatic agent is a minor groove binder, AEB, ~~or~~ AEVB, MMAF, MMAE, or AFP.

38. **(Currently Amended)** The method of claim 34, wherein ~~the drug is MMAF, MMAE, or AFP~~ at least two drugs are conjugated to the antibody and each drug is conjugated to one interchain thiol.

39. **(Original)** The method of claim 34, wherein the reducing agent is DTT or TCEP.

40-42. **(Canceled)**

43. **(Currently Amended)** The method of ~~claims 40~~ claim 34, further comprising purifying the partially reoxidized antibody.

44. **(Canceled)**

45. **(Currently Amended)** A method of reducing antibody interchain disulfide bonds and conjugating a drug to the resulting interchain thiols to selectively locate the drug ~~drugs~~ on the antibody, comprising:

partially reducing the antibody with a reducing agent to form at least two interchain thiols; and

conjugating the drug to ~~the one~~ interchain thiols thiol of the partially reduced antibody.

46. **(Original)** The method of claim 45,  
wherein the antibody is partially reduced with a limiting concentration of a reducing agent in a buffer with a chelating agent; and

wherein the drug is conjugated by cooling the antibody solution and dissolving the drug in a cold solvent and mixing with the antibody solution;

allowing the antibody and drug solution to incubate for a period of time sufficient to form an antibody-drug conjugate;

quenching the excess drug with a thiol-containing reagent; and  
purifying the resulting conjugate.

47. **(Currently Amended)** The method of claim ~~[[ 46]]~~45, wherein ~~the antibody is partially reduced for about 1 hour at about 37°C at least two drugs are conjugated to the resulting interchain thiols and each drug is conjugated to one interchain thiol.~~

48-49. **(Canceled)**

50. **(Currently Amended)** The method of claim ~~45~~46, wherein the thiol-containing reagent is cysteine or N-acetyl cysteine.

51. **(Original)** The method of claim 45, wherein the reducing agent is DTT or TCEP.

52. **(Original)** The method of claim 46, wherein the buffer is a sodium borate solution and the chelating agent is diethylenetriaminepentaacetic acid.

53. **(Original)** The method of claim 46, wherein the chelating agent is ethylenetriaminepentaacetic acid or EDTA.

54. **(Original)** The method of claim 45, further comprising purifying the reduced antibody.

55-57. **(Canceled)**

58. **(Original)** The method of claim 45, wherein the reduced antibody is not purified after partial reduction and prior to conjugation.

59-61. **(Canceled)**

62. **(Currently Amended)** The method of claim ~~45~~46, wherein the solvent is acetonitrile, alcohol or DMSO.

63. **(Original)** The method of claim 45, wherein the drug is a cytotoxic or a cytostatic agent.

64. **(Currently Amended)** A method of producing an antibody with selective conjugation of a drug comprising:

fully reducing the antibody for a period of time sufficient to produce interchain thiols, as determined by DTNB titration, by adding a large excess of a reducing agent and incubating the solution at about 37 °C for about 30 minutes;

purifying the antibody;

partially reoxidizing the antibody using an oxidizing agent to form at least one interchain disulfide bond by

cooling the reduced antibody to 0 °C;

treating the reduced and cooled antibody with 1.5 to 2.5 molar equivalents of the oxidizing agent;

mixing the solution by inversion;

allowing the solution to incubate at about 0 °C for about 10 minutes;

purifying the partially reoxidized antibody;

conjugating the drug to ~~the~~ one interchain ~~thiols~~ thiol of the partially reoxidized antibody to form a conjugated antibody; and

purifying the conjugated antibody.

65. **(Currently Amended)** The method of claim 64, wherein ~~the reducing agent is DTT or TCEP~~ at least two drugs are conjugated to the antibody and each drug is conjugated to one interchain thiol.

66-72. **(Canceled)**

73. **(Currently Amended)** A method of preparing a conjugate of a protein having one or more disulfide bonds and a drug, ~~and a drug reactive with free thiols, which comprises~~ comprising:

partially reducing the protein with a reducing agent; and

conjugating the drug reactive with free thiols to the partially reduced protein.

74. **(Currently Amended)** A method of preparing a conjugate of a protein having one or more disulfide bonds and a drug, ~~and a drug reactive with free thiols, which comprises comprising:~~

- fully reducing the protein with a reducing agent;
- partially reoxidizing the protein with a reoxidizing agent; and
- conjugating the drug reactive with free thiols to the ~~antibody protein~~.

75. **(Currently Amended)** A method of forming a ~~partially loaded an~~ antibody ~~that is partially loaded with a drug, comprising:~~ comprising:

- providing a solution containing an ~~antibody~~ antibody;
- adjusting the pH of the antibody solution to about pH 7.5 and adding a chelating agent;

- heating the antibody solution to about 37 °C;
- adding a molar excess of TCEP to the antibody solution and reacting for a sufficient period of time at about 37 °C to partially reduce the interchain disulfide groups of the antibody to form interchain thiols;

- cooling the antibody solution to between about 2-8 °C;
- conjugating the drug to ~~the one~~ interchain ~~thiols~~ thiol of the partially reduced antibody by adding a slight molar excess of the drug to the antibody solution and reacting for a sufficient period of time to form the partially loaded antibody; and
- purifying the partially loaded antibody.

76. **(New)** A partially loaded, modified protein, comprising:

- a binding region for interaction with a binding partner;
- at least two points of conjugation having a similar accessibility or activability for conjugation of a drug or label by chemical means;
- at least two drugs or labels, each drug or label covalently linked to one point of conjugation;
- wherein less than all of the possible points of conjugation having a similar accessibility or activability are linked to a drug or label.

77. (New) The modified protein of claim 76, wherein the protein comprises an antibody, a receptor, a receptor ligand, a hormone, or a cytokine.

78. (New) The modified protein of claim 76, wherein the points of conjugation are thiol groups, amino groups, vicinal hydroxyl groups, hydroxyl groups, or carboxyl groups.

79. (New) The modified protein of claim 77, wherein:  
the protein is an antibody;  
the binding region is an antigen-binding domain of the antibody;  
the points of conjugation are thiol groups; and  
the antibody comprises at least one interchain disulfide bond.

80. (New) The modified protein of claim 79, wherein a thiol group is a thiol group of a cysteine residue.

81. (New) The modified protein of claim 80, wherein the thiol group is a thiol group of a cysteine residue of an interchain disulfide bond.

82. (New) The modified protein of claim 81, wherein each drug or label is conjugated to a thiol group of a cysteine residue of an interchain disulfide bond.

83. (New) The modified protein of claim 79, wherein at least one thiol group is formed by alkylation of the epsilon amino group of a lysine residue.

84. (New) The modified protein of claim 76, wherein two drugs are conjugated to the modified protein.

85. (New) The modified protein of claim 79, wherein two drugs are conjugated to the modified protein.

86. (New) The modified protein of claim 76, wherein the drug is a cytotoxic or cytostatic drug or an immunosuppressive agent.

87. (New) The modified protein of claim 79, wherein the drug is a cytotoxic or cytostatic drug or an immunosuppressive agent.

88. (New) The modified protein of claim 87, wherein the antibody comprises at least four cytotoxic or cytostatic drugs, each drug conjugated to an interchain thiol.

89. (New) The modified protein of claim 79, wherein the conjugated antibody has the configuration of species 4A, 4B, 4C, 4D, 4E, or 4F of Figure 1.

90. (New) The modified protein of claim 79, wherein the antibody is a humanized or chimeric antibody.

91. (New) The modified protein of claim 87, wherein the drug is AEB, AEVB, MMAF, MMAE, or AFP.

92. (New) A composition of modified antibodies, comprising:  
at least two species of th modified antibody of claim 79, wherein the species are selected from species 4A, 4B, 4C, 4D, 4E or 4F of Figure 1.

93. (New) A pharmaceutical composition comprising the modified protein of claim 76 and a pharmaceutically acceptable carrier.

94. (New) A pharmaceutical composition comprising the modified protein of claim 79 and a pharmaceutically acceptable carrier.

95. (New) The pharmaceutical composition of claim 94 wherein a thiol group is a thiol group of a cysteine residue.

96. (New) The pharmaceutical composition of claim 95, wherein the thiol group is a thiol group of a cysteine residue of an interchain disulfide bond.

97. (New) The pharmaceutical composition of claim 96, wherein each drug or label is conjugated to a thiol group of a cysteine residue of an interchain disulfide bond.

98. (New) The pharmaceutical composition of claim 94, wherein a thiol group is formed by alkylation of the epsilon amino group of a lysine residue.

99. (New) The pharmaceutical composition of claim 94, wherein two drugs are conjugated to the modified protein.

100. **(New)** The pharmaceutical composition of claim 94, wherein the drug is a cytotoxic or cytostatic drug or an immunosuppressive agent.

101. **(New)** The pharmaceutical composition of claim 100, wherein the antibody comprises at least four cytotoxic or cytostatic drugs, each drug conjugated to an interchain thiol.

102. **(New)** The pharmaceutical composition of claim 94, wherein the conjugated antibody has the configuration of species 4A or 4B, 4C or 4D, 4E, or 4F of Figure 1.

103. **(New)** The pharmaceutical composition of claim 94, wherein the antibody is a humanized or chimeric antibody.

104. **(New)** The pharmaceutical composition of claim 100, wherein the drug is AEB, AEVB, MMAF, MMAE, or AFP.

105. **(New)** The pharmaceutical composition of claim 94, wherein there is an average of 4 drugs per antibody.

106. **(New)** The pharmaceutical composition of claim 94, wherein there is an average of 2 drugs per antibody.

107. **(New)** A method for the treatment of cancer, immune disease, autoimmune disease or infectious disease in a patient, comprising administering to the patient an amount of the modified protein of claim 10.

108. **(New)** A method for the treatment of cancer, immune disease, autoimmune disease or infectious disease in a patient, comprising administering to the patient an amount of the modified protein of claim 76.

109. **(New)** The method of claim 108 wherein:  
the protein is an antibody;  
the binding region is an antigen-binding domain of the antibody;  
the points of conjugation are thiol groups; and  
the antibody comprises at least one interchain disulfide bond.



110. (New) A method for the treatment of cancer, immune disease, autoimmune disease or infectious disease in a patient, comprising administering to the patient an amount of the pharmaceutical composition of claim 93.

111. (New) The method of claim 110 wherein;  
the protein is an antibody;  
the binding region is an antigen-binding domain of the antibody;  
the points of conjugation are thiol groups; and  
the antibody comprises at least one interchain disulfide bond.

112. (New) The method of claim 75, wherein at least two drugs are conjugated to the antibody and each drug is conjugated to one interchain thiol.